

Cervical sampling for diagnosis of genital chlamydial infection with a new brush device

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Abstract

Objectives—to compare a new sampling device, a brush, Accellone-Multi-Instrument (AMI), with a dacron-tipped swab for detection of *Chlamydia trachomatis* in endocervical specimens, and to evaluate if consecutive multiple cervical sampling as compared with such a single specimen would increase the sensitivity.

Methods—501 females attending an STD clinic and 172 females attending a family planning clinic were examined prospectively. Two cervical specimens were collected from each woman. *C trachomatis* were detected by culture or enzyme immunoassay (IDEIA-III). Positive EIA samples were confirmed by a direct immunofluorescent test.

Results—When cervical specimens were collected with the brush as the first device, 92% of the culture-positive cases were detected, and when the samples were collected with the dacron-tipped-swab first, 84% of the culture-positive cases were detected ($p < 0.05$). The first collected specimen detected 89% of the culture-positive cases and 81% of those that were positive by IDEIA.

Conclusions—The study indicates that the AMI brush is superior to non-toxic, dacron-tipped swabs for detection of *C trachomatis* in cervical samples by cell culture but not by ELISA, and that the sensitivity could be improved by analysing multiple cervical samples.

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Introduction

The means to detect genital infection by *C trachomatis* is still not optimal. Thus it has been estimated that only approximately 70-80% of infected cases will be detected by presently available diagnostic techniques.¹⁻²

A number of factors can influence the likelihood of detecting the organism in clinical samples, such as the type of specimen analysed (cervical, urethral, voided urine etc), the sampling device and sampling technique used, the storage, and transport conditions as well as the laboratory analytical technique used.^{1,3}

The purpose of this study was to compare a new synthetic sampling brush, the Accellone-Multi-Instrument, with a dacron-tipped swab. Furthermore, to evaluate if consecutive multiple cervical sampling as

compared with such a single specimen would increase the detection rate for *C trachomatis* from the cervix when analysed either with cell culture or ELISA.

Material and methods

Study populations

Specimens were collected from 501 females attending a sexually transmitted disease clinic (Group A), who were aged 18-35 years (mean 25.0), many of whom had symptoms. Also included in the study were 172 asymptomatic females (Group B) attending a family planning clinic for contraceptive advice, between the ages of 16-35 years (mean age 23.4 years).

Specimen collection

Two cervical specimens were collected from each woman, one specimen with a new type of brush, the Accellone-Multi-Instrument® (AMI) (Medscand, Malmö, Sweden) and the another specimen with a dacron-tipped swab. The former is a brush with a thin plastic shaft with its tip covered with soft bristles.

Before taking the samples the exocervix was cleansed with a dry cotton pad. On even calendar dates the dacron-tipped swab was used first and vice versa on odd calendar dates.

The specimens were placed in the transport medium supplied by the kit procedure (Novo Bio Labs) of the enzyme immunoassay used or in a sucrose-phosphate (2-SP) buffer for tissue culture studies.

Detection of chlamydia trachomatis

Within 72 hours of sampling, samples from Group A were inoculated into cycloheximide-treated McCoy cell cultures.

Samples from Group B were analysed in an enzyme immunoassay, (IDEIA-III (Novo Bio Labs)). Positive samples were confirmed by a direct immunofluorescence test (DIF), (MicroTrak (Syva)). Only samples positive either by culture or in both the IDEIA and the DIF tests were regarded as true positives.

Results

Of the 673 women included in the study, 85 (12.6%) were found to be infected by *C trachomatis*. Of the 501 females in Group A, 64 (12.8%) were positive as evidenced by culture. Of the 501 samples collected by use of a dacron-tipped swab, 51 (10.2%) were culture-positive. They constituted 79.7%

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Table 1 Comparison of a brush sampling device (Accellone-Multi-Instrument) and a dacron-tipped swab for detection of *Chlamydia trachomatis* by culture in group A (N = 501).

Sampling device	Culture test			
Brush	+	+	-	-
Swab	+	-	+	-
Total	46	13	5	437

In total, 59 and 51 of the chlamydia positive specimens had been sampled by the brush and the dacron-tipped swab, respectively.

of the cases detected to be infected with *C trachomatis*.

With the AMI brush, 59 (11.8%) were culture-positive, i.e. 92.2% of all chlamydia cases detected in Group A (table 1).

In Group B, 21 (12.2%) of the 172 women were chlamydia-positive in IDEIA-111 in all cases confirmed by DIF. Both the dacron-tipped swab and the AMI brush revealed 17 (9.9%) of the chlamydia-infected cases which were 81% of those found to be infected in Group B (table 2).

When samples were taken by the dacron-tipped swab before the brush in Group A, 21 of 25 (84%) chlamydia-positive cases were detected by the first specimens, while when the AMI brush was the first device used, 36 of 39 (92%) specimens were culture-positive ($p < 0.05$) (table 3).

In Group B, 11 specimens were ELISA-positive when the dacron-tipped swab was the first device used, while this was true for 10 specimens when the AMI brush was used first (table 4).

The first collected specimens detected 86% (57/66) of the culture-positive cases and 81% (17/21) of those who were positive by IDEIA-111.

No samples collected with the AMI brush or the dacron-tipped swab were found to be cytotoxic.

Traces of blood were observed in 297 (44%) of the samples collected by the AMI device and in 152 (23%) of those collected by the dacron-tipped swab.

Discussion

The detection of the intracellular organism *C trachomatis* in tissue cell culture is related to the quantity of epithelial cells in the specimen collected.¹

The detection rate is dependent on the amount of chlamydia organisms (or antigen) present in the specimen and the toxicity of

Table 2 Comparison of a brush sampling device (Accellone-Multi-Instrument) and a dacron-tipped swab used for detection of *Chlamydia trachomatis* by immunofluorescent-confirmed enzyme immunoassay (IDEIA-111) (N = 172).

Sampling device	Immunoassay			
Brush	+	+	-	-
Swab	+	-	+	-
Total	13	4	4	151

In total, 17 chlamydia-positive specimens were detected in samples collected both by the brush and the dacron-tipped swab.

Table 3 Comparison of detection rate of *Chlamydia trachomatis* by culture between first and second consecutive sampling occasion in women sampled by a brush device (Accellone-Multi-Instrument) and a dacron-tipped swab. (N = 501)

Sampling device	Sampling order	Result			
Brush	1	+	+	-	-
Swab	2	+	-	+	-
Total		27	9	3	209
Swab	1	+	+	-	-
Brush	2	+	-	+	-
Total		19	2	4	228

In total, 36 women were chlamydia-positive in the first sampling from the cervix when the sampling was made with the brush, while 21 women were positive when the dacron-tipped swab was first to be used ($p < 0.05$).

the sampling device is a factor which influences the outcome of culture studies.³ The recovery rate has been reported to be improved by using a brush device, the Cytobrush® compared with dacron- or cotton-tipped swabs.⁴⁻⁷ The brushes yield an increased number of epithelial cells as compared with ordinary swabs in samples collected from the cervical channel.⁸ The same is true for the number of elementary bodies (EB) of *C trachomatis* detected in such samples analysed by DIF. However, the advantage with the brush device has not been confirmed in other studies.⁸⁻¹³

The brush can induce bleeding from the friable cervical channel¹¹ and a cytotoxic effect on McCoy cells, when samples have been contaminated with blood, has been reported. Bleeding occurred in 44% of the the sampled cases using the AMI-brush and in 23% of the cases using the dacron-tipped swab. The bleeding did not seem to affect the detection of *C trachomatis*.

However, samples taken with the AMI-brush has a higher sensitivity for detection of *C trachomatis* by tissue cell cultures than when dacron-tipped swabs were used, which may be explained by the former sampling result in a higher number of epithelial cells in the specimen.

Dunlop *et al*¹⁴ found that multiple endocervical swabs increased the sensitivity of detecting genital infection by *C trachomatis*. They found that the first collected swab iden-

Table 4 Comparison of the detection rate of *Chlamydia trachomatis* between first and second consecutive sampling occasion from the cervix in specimens collected by a brush device (Accellone-Multi-Instrument®) and a dacron-tipped swab when analysed by IDEIA-111 and confirmed by direct immunofluorescence (Mikro Trak) (N = 172).

Sampling device	Sampling order	Result			
Brush	1	+	+	-	-
Swab	2	+	-	+	-
Total		6	2	2	77
Swab	1	+	+	-	-
Brush	2	+	-	+	-
Total		7	2	2	74

In total, 8 women were chlamydia-positive when the first sampling from the cervix was made with the brush, while 9 women were positive when the dacron-tipped swab was first to be used. (Not significant).

tified 73%, the second an additional 17%, and the third swab still 10% more culture-positive women. They concluded that in culture studies, samples obtained by use of a single swab underestimate the prevalence of genital chlamydial infection. In other studies^{10 15}, only a marginal difference by multiple and single sampling has been demonstrated.

In the present study only 89% of the chlamydia-infected women in group A and only 81% of those in group B were detected with aid of the first collected specimen. That is, about 10–20% of the infected cases, would have been missed by culture and twice as many would not have been detected in ELISA if only a single sample had been obtained.

Sampling from the urethra in addition to the cervix in asymptomatic women, add less than 10% of chlamydia-positive cases as compared with when only cervical sampling is made.^{14 16} Urethral sampling is, however, often painful and may cause urethritis symptoms because of injury of the urethral mucosa. Two consecutive cervical swabs would be effective in diagnosing genital chlamydial infection without being painful. Thus, two consecutive samples added 11 (15%) positive cases as compared with when only one cervical sample (collected by the aid of the dacron-tipped swab or the AMI device) was analysed.

There was no difference in the subjective feeling of discomfort after sampling with the AMI and the dacron-tipped swab.

To sum up, our study demonstrated that the Accellone-Multi-Instrument is equal or superior to non-toxic dacron-tipped swabs for detection of *C. trachomatis* by cell culture of endocervical specimens and that duplicate endocervical specimens increases the sensitivity both when analysing the samples by culture and EIA in cervical samples.

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